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Prevalence and antibiotic resistance of commensal Streptococcus pneumoniae in nine European countries

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ABSTRACT

The human microbiota represents an important reservoir of antibiotic resistance. Moreover, the majority of antibiotics are prescribed in primary care. For this reason, we assessed the prevalence and antibiotic resistance of nasal carriage strains of Streptococcus pneumoniae, the most prevalent bacterial causative agent of community-acquired respiratory tract infections, in outpatients in nine European countries. Nasal swabs were collected between October 2010 and May 2011, from 32,770 patients, recruited by general practices in nine European countries. Overall prevalence of S. pneumoniae nasal carriage in the nine countries was 2.9%. The carriage was higher in men (3.7%) than in women (2.7%). Children (4–9 years) had a higher carriage prevalence (27.2%) compared with those older than 10 years (1.9%). The highest resistance observed was to cefaclor. The highest prevalence of multidrug resistance was found in Spain and the lowest prevalence was observed in Sweden.

Streptococcus pneumoniae is an important causative agent of community-acquired pneumonia, otitis media, bacteremia and meningitis, especially in immunocompromised patients, young children and elderly individuals [1]. The upper respiratory tract is the main ecological niche of S. pneumoniae[2]. The colonization rates range, depending on age, between 3 and 60% [3,4] and depend on geographic area, age, genetic background and socioeconomic status of the host [1].

The introduction of pneumococcal conjugate vaccines has led to a major decrease in pneumococcal carriage and the prevalence of pneumococcal-related diseases [5]. Depending on the vaccine used, emergence of new colonizing serotypes has been observed that have replaced the vaccine serotypes [6,7]. Many host, environment and microbiological factors play a role in the increase of susceptibility to carry S. pneumoniae. One example of the microbiological factors is coinfection with influenza virus. Several reports have observed pneumococcal transmission via

contact of a nonpneumococcal carrier with infected respiratory secretion of an influenza infected pneumococcal carrier [8], or by person-to-person contact [9]. Most population studies conducted thus far have focused on nasopharyngeal carriage in children [10], on the prevalence of invasive isolates [1,5,7] and on the effectiveness of vaccination strategies [6,11–14]. Pneumococcal carriage in adults has hardly been described, despite the fact that colonization is essential to understand bacterial epidemiology and to assess the effectiveness of preventive strategies [15]. During the last few decades, antimicrobial drug resistance has increased worldwide [16]. The human microbiota is generally considered as the main reservoir for antibiotic resistant genes and resistant microorganisms [17,18]. The acquisition of antibiotic resistance in this environment is enhanced by the enormous potential of genomic exchange and mutations [19].

One of the aims of the APRES study, was to assess the prevalence and antibiotic resistance of nasal carriage of S. aureus and S. pneumoniae in general practice patients and to compare antibiotic resistance patterns of these isolates [20] with different antibiotic prescription policies. The results ofS. aureus analyses have been described before [21]. Here, we report the results of the prevalence and antibiotic resistance of S. pneumoniae carriage in the participating countries.

MATERIALS & METHODS

• Patient population

The study design was described previously by van Bijnen et al. [20]. In short, 20 general practitioners (GP) were recruited per country, by national GP networks in nine European countries (Austria, Belgium, Croatia, France Hungary, Spain, Sweden, The Netherlands and the UK), and aimed to each provide 200 nasal swabs, from patients older than 4 years, with the exception of the UK where patients were 18 years or older (due to ethical committee constraints). To be included in the study, patients should have visited the practice for a noninfectious condition, not used antibiotics nor been hospitalized in the 3 months preceding the swab collection. Immunocompromised patients, as well as nursing home residents, were excluded. Included patients were sampled consecutively.

Information of the participating patients was derived from a questionnaire and included age, gender, profession (livestock farming, working in healthcare or in day care/nursery school), the number of GP visits during the last year, living with children younger than 5 years (other than the index patient), suffering from a chronic skin condition and vaccination against pneumococci and/or influenza (season 2009–2010 and/or 2010–2011). Consent and information of children were obtained from the parents or the legal representative. The questionnaire did not differentiate between 2009 and 2010 seasonal and pandemic vaccines. Information on pneumococcal and influenza vaccination policy in each participating country was obtained from a literature search.

Prior to the start of the study, GPs received a protocol for sampling the nasal swabs [21]. Isolation and identification of S. pneumoniae was performed in a national microbiological laboratory in each participating country except for France. These swabs were analyzed in the microbiological laboratory of Maastricht University Medical Center ([MUMC], The Netherlands), because no laboratory in France was prepared to perform these analyses. All (putative) pneumococci isolated at the national laboratories were kept frozen in skimmed milk at -80oC and sent at the end

of the study period on dry ice to the laboratory in Maastricht for antibiotic susceptibility testing.
The antibiotic resistance of the isolates was performed with microdilution for cefuroxime, cefaclor, ceftazidime, clarithromycin, clindamycin, ciprofloxacin, moxifloxacin, penicillin, tetracycline and trimethroprim-sulfamethoxazole. The control strain was S. pneumoniae ATCC 49619. Invalid results due to nonhomogenous growth were repeated twice. The method used was in accordance with EUCAST guidelines and EUCAST epidemiological cutoffs were used as resistance breakpoints [22].
Patients with incomplete demographic information or laboratory data, as well as erroneous samples (no bacterial growth noted at laboratory analysis), were excluded from the analyses.

- **Statistical analysis**

Results were analyzed using the PASW software package 19.0. A p-value of <0.05 was considered as statistically significant.
To determine pneumococcal carriage determinants odd ratios (OR)s were calculated. OR was defined as the cross-product ratio of the numbers shown in the two-by-two contingency table of flu vaccination, pneumococcal vaccination, occupation, having a chronic skin disease and living with other young children. The total number of patients who answered the specific question was used as the denominator.
To account for nonrandom clustering of our data at family level and to control whether age and gender affect the pneumococcal carriage prevalence, a three level multilevel logistic regression model (country, GP and patient) was estimated using MLN-Software for N-level analysis package.
Multidrug resistance (MDR) was defined as being resistant to three or more classes of antibiotics.
The statistical analysis was made on two age groups: <10 years and ≥ 10 years. This cutoff was used to differentiate young children versus the older ones and adults as after 10 years of age pneumococcal carriage was stable [1].

- **Role of the funding source**

The sponsor of this study had no role in study design, data collection, data analysis data interpretation or writing of this report. The corresponding author had full access to all the data on the study and had final responsibility for the submission for publication, following agreement from all authors.

RESULTS

The number of participants ranged between 3132 in Belgium and 4017 in Hungary, resulting in a total of 32,770 patients recruited (Table 1). Patients who did not meet the inclusion criteria because of their demographic characteristics, or incorrect sampling were excluded from the analysis [21] resulting in a total of 32,161 patients available for the data analysis.

[TABLE 1]

- **Prevalence of nasal carriage**

The overall crude prevalence of S. pneumoniae nasal carriage was 2.9% (n = 937), and ranged from 1.2% in Austria (n = 38) and the UK (n = 36) to 4.4% in France (n = 170).
The prevalence of S. pneumoniae carriage was higher in men (3.7%; 95% CI: 3.4–4.0) than in women (2.7%; 95% CI: 2.1–2.5). Children (4–9 years) had a higher carriage prevalence (27.2%; 95% CI: 24.7–29.6) compared with those older than 10 years (1.9%; 1.8–2.1). The highest carriage rate in the 4–9 year old was in Spain (24.0%) followed by France (22.2%) (Table 2).

- **Vaccination rates**

Immunization strategies against S. pneumoniae and seasonal influenza and pandemic influenza A virus differed between the nine participating countries. Variation in pneumococcal immunization included vaccination strategy (recommended or voluntary, universal or risk groups only), vaccine type, vaccination schedule, administrated doses and targeted groups.
Seasonal influenza and pandemic Flu A immunization differed in targeted groups and clinical risk indications. Spain had the highest overall pneumococcal vaccination rate with 29.2% (46.4% in children younger than 9 years, and 28.3% in adolescent and adults), whereas Croatia had the lowest one with an overall rate of 0.7% (3.2% in patients younger than 9 years and 0.6% in patients of 10 years or older). France had the highest vaccination rate in children <9 years old (46.7%). Overall, 11.6% of all participating patients were vaccinated (Table 3).

- **Determinants of nasal carriage**

Living with children younger than 5 years of age (or other children if the individual is a child <5 years old), resulted in a four-fold increased risk to carry S. pneumoniae (OR: 3.92; 3.41–4.50). In all participating countries, patients vaccinated against S. pneumoniae had a (nonsignificant) higher prevalence of pneumococcal carriage. The carriage prevalence among the vaccinated group was 3.5%, whereas in the unvaccinated patients this was 2.9% (OR: 1.2; 0.99–1.48). Seasonal and pandemic influenza A vaccination had no effect on the nasal pneumococcal carriage.

[TABLE 2] [TABLE 3]

- **Antibiotic resistance**

Antibiotic resistance results were available for 928 (99.0%) of 937 S. pneumoniae isolates.
Resistance to cefaclor was most common: overall 52.3% and ranging between 30.6% in the UK and 77.3% in Belgium. No resistance was observed for moxifloxacin and ciprofloxacin. Tetracycline showed the highest intercountries variability ranging between 1.9% in Sweden and 29.9% in Spain (Table 4). The most frequently used antibiotics in outpatients were penicillin, macrolides and tetracycline [14]. These antibiotics showed a resistance that ranged between 3.9% (Sweden) and 31.7% (Spain) for penicillin, 1.9% (Sweden) and 29.3% (Spain) for clarythromycin, and 1.9% (Sweden) and 29.9% (Spain) for tetracycline (Table 5).

- **MDR**

Spain had the highest prevalence of MDR (resistance for three or more classes), in other words, 50.4%, (n = 63). The lowest percentage was observed in Sweden with 3.8% (n = 2). Thirty isolates were resistant to six classes, of which ten were from Croatia. Overall, 27 of these 30 isolates were resistant to all tested antibiotics, except for moxifloxacin and ciprofloxacin and three were resistant to all tested antibiotics. The most prevalent MDR was resistance to five classes (n = 70). The most common combination was resistance to cephalosporines (cefaclor, cefuroxime and ceftazidime), macrolides fluoroquinolone, penicillin and tetracycline (n = 40). Most of these isolates were isolated in France (n = 17) and Spain (n = 16).

[TABLE 4] [TABLE 5]

DISCUSSION

In this study, nasal samples from more than 32,000 outpatients from nine European countries were collected and analyzed. The overall S. pneumoniae carriage prevalence was 2.9% and resistance to cefaclor was the most common among the isolates with 52.3%. Of all resistant isolates, 19.4% were nonsusceptible to penicillin. MDR was observed in 33.9%, ranging from 3.8% in Sweden to 50.4% in Spain.

To the best of our knowledge, our study is the first to compare the prevalence and antibiotic resistance of commensal S. pneumoniae in healthy outpatients aged 4+ in several European countries. For this aim, existing GP networks were involved for patient recruitment. In addition, standardized and validated protocols were used to prevent inter laboratory differences.

Our study has some limitations. For logistic reasons, in other words, feasibility to perform a study in nine different countries with 20 different general practitioners we choose to take nasal swabs instead of nasopharyngeal ones. In all swabs, even in the nasal swabs from France that were sent to Maastricht, the prevalence was in accordance to the expected one [23]. Furthermore, based on these observations, we may conclude that taken nasal swabs do not influence the results negatively. Another limitation is that, due to ethical constraints, the lower age limit for eight countries was 4 years of age, for the UK 18 years. This hampers the assessment of pneumococcal carriage and the antibiotic resistance in the lowest age group (<4 years). Finally, we did not collect clinical data, such as among others comorbidities and recent hospitalization.

A wide variation in pneumococcal carriage prevalence was found among the participating countries. One of the major factors that influence pneumococcal colonization is the vaccination strategy, including vaccine type, whether vaccine was given to all children >2 years, universal or restricted to clinical risk groups only, such as patients suffering from chronic respiratory disease or cardiovascular disease (risk based), if the implementation of a vaccine was followed by a catch-up pneumococcal vaccination and the number of doses given. The high carriage prevalence in Sweden might be explained by the late introduction of vaccination to the national immunization program (2009). The introduction of the vaccine without a catch up campaign [24] does not allow individuals who exceeded the immunization age to still be vaccinated. In Croatia and Spain pneumococcal vaccine was given only to patients belonging to specific risk groups. This strategy reduces the number of vaccinated

individuals and might increase the number of colonized patients. In Belgium, Croatia and The Netherlands, only PCV-7 vaccine is used [24]. The emergence of nonvaccine serotypes after the introduction of this vaccine (replacement phenomenon) might explain the high prevalence of pneumococcal carriage in these countries [5].

No significant difference was noticed between carriage prevalence in vaccinated and unvaccinated individuals. Our results support earlier studies with comparable outcomes. In a carriage study carried on 683 subjects from 217 households in England and Wales, van Hoek et al. found no change in pneumococcal carriage and a decrease of PCV7 vaccine serotypes transmission in vaccinated and unvaccinated individuals 6 years after the introduction of PCV7 vaccine, compared with carriage prevalence before the introduction of this vaccine [25]. In another carriage study, performed by Principi et al., enrolling 2076 children and adolescents from Italy, it was found that pneumococcal prevalence was higher in vaccinated individuals than in unvaccinated ones [26]. Moreover, they found that the prevalence vaccine serotypes was comparable to this nonvaccine serotypes, suggesting no role of serotypes replacement. Taken together with these results, our results might be explained by the emergence of none-vaccine serotypes that cocirculated but were suppressed by vaccine serotypes. Another explanation is the possibility that the protection effect of vaccination against pneumococcal carriage might decrease over the time, allowing a re-emergence of vaccine serotypes in vaccinated patients.

In two separate mice model studies, researchers suggested that influenza virus infection enhanced the carriage of S. pneumoniae by an increase in pneumococcal load in carriers and the increase of susceptibility in influenza infected cohoused subjects [27,28]. This suggests that influenza vaccination, by reducing the spread of infected droplets, might reduce pneumococcal transmission and, consequently, pneumococcal carriage. However, our data did not confirm this suggestion.

In this study, a large variability was found between the participating countries in resistance prevalence to the commonly used antimicrobial agents in outpatients (penicillin, macrolide and tetracycline).

Belgium, France, Spain and Croatia that in 2010 had a penicillin consumption of 16.3, 15.6, 12.6 and 9.5 defined daily dose (DDD) per 1000 inhabitants per day, respectively [29], had the highest rate of antibiotic resistance to this antibiotic class. The Netherlands, with a penicillin consumption of 4.4 DDD per 1000 inhabitants per day [29], had one of the lowest penicillin resistance. In Sweden, despite the higher rate of penicillin consumption compared with The Netherlands (7.1 DDD per 1000 inhabitants per day) [29], the resistance rate to this agent was the lowest. This might be explained by the use in Sweden of narrow spectrum penicillins (more than 60% of the used penicillins). Furthermore, our data showed a high resistance to clarithromycin in Croatia. The consumption of this agent represents, with roxithromycin, more than 30% of the macrolides, lincosamides and streptogramin use in Croatia [30].

Differences in antibiotic consumption between the participating countries might not be the only explanation of the resistance rate variations. In fact, Spain with tetracycline consumption of 0.7 DDD per 1000 inhabitants per day [29], has the highest resistance rate, whereas Sweden with a more than four-fold consumption (3.3 DDD per 1000 inhabitants per day) [29], had the lowest resistance prevalence. These results underscore that other factors than antibiotic pressure, socioeconomic,

behavioral and cultural determinants and differences in healthcare policies among the participating countries [31], as well as clonal spread of resistant microorganisms [32] and antibiotic cross-resistance between members of antibiotic classes [33] might influence antibiotic resistance prevalence.

Our study highlights the prevalence of antibiotic resistant S. pneumoniae carriage strains in the outpatient setting in nine European countries. These data are important to make a rational empiric choice and to create antibiotic prescription guidelines for the treatment of S. pneumoniae by GPs (i.e., the outpatient setting). As the majority of human antibiotic use is prescribed outside healthcare settings [16,34], optimal use of antibiotics based on actual antibiotic resistance data is important to support the control of antibiotic resistance problem. Professionals involved in the set-up/reviewing of existing national antibiotic prescription guidelines have to take into account the data presented of the country involved.

Conclusion

A low S. pneumoniae nasal carriage prevalence was assessed in general practice patients in nine European countries. A large variability in antibiotic resistance was noted among the participating countries, with the highest overall resistance in Spain and the lowest in Sweden.

Financial & competing interests disclosure

The authors have no relevant affiliations or financial involvement with any organization or entity with a financial interest in or financial conflict with the subject matter or materials discussed in the manuscript. This includes employment, consultancies, honoraria, stock ownership or options, expert testimony, grants or patents received or pending, or royalties.

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Ethical conduct of research

The authors state that they have obtained appropriate institutional review board approval or have followed the principles outlined in the Declaration of Helsinki for all human or animal experimental investigations. In addition, for investigations involving human subjects, informed consent has been obtained from the participants involved.

References

1. Bogaert D, De Groot R, Hermans PW. Streptococcus pneumoniae colonisation: the key to pneumococcal disease. Lancet Infect. Dis. 4(3), 144–154 (2004). [CrossRef] [Medline] [CAS]
2. Austrian R. Some aspects of the pneumococcal carrier state. J. Antimicrob. Chemother.18(Suppl. A), 35–45 (1986). [Medline]
3. Dagan R, Melamed R, Muallem M et al. Nasopharyngeal colonization in southern Israel with antibiotic-resistant pneumococci during the first 2 years of life: relation to serotypes likely to be included in pneumococcal conjugate vaccines. J. Infect. Dis. 174(6), 1352–1355 (1996).[CrossRef] [Medline] [CAS]
4. Cardozo DM, Nascimento-Carvalho CM, Andrade AL et al. Prevalence and risk factors for nasopharyngeal carriage of Streptococcus pneumoniae among adolescents. J. Med. Microbiol.57(Pt 2), 185–189 (2008). [CrossRef] [Medline]

5. Leibovitz E. The effect of vaccination on Streptococcus pneumoniae resistance. Curr. Infect. Dis. Rep. 10(3), 182–191 (2008). [CrossRef] [Medline]
6. Spijkerman J, Prevaes SM, Van Gils EJ et al. Long-term effects of pneumococcal conjugate vaccine on nasopharyngeal carriage of S. pneumoniae, S. aureus, H. influenzae and M. catarrhalis. PLoS ONE 7(6), e39730 (2012). [CrossRef] [Medline] [CAS]
7. Mehr S, Wood N. Streptococcus pneumoniae – a review of carriage, infection, serotype replacement and vaccination. Paediatr. Respir. Rev. 13(4), 258–264 (2012). [CrossRef][Medline]
8. Kadioglu A, Weiser JN, Paton JC et al. The role of Streptococcus pneumoniae virulence factors in host respiratory colonization and disease. Nat. Rev. Microbiol. 6(4), 288–301 (2008).[CrossRef] [Medline] [CAS]
9. Brundage JF. Interactions between influenza and bacterial respiratory pathogens: implications for pandemic preparedness. Lancet Infect. Dis. 6(5), 303–312 (2006). [CrossRef] [Medline]
10. O'brien KL, Nohynek H. Report from a WHO Working Group: standard method for detecting upper respiratory carriage of Streptococcus pneumoniae. Pediatr. Infect. Dis. J. 22(2), e1–e11(2003). [CrossRef] [Medline]
11. Weil-Olivier C, Van Der Linden M, De Schutter I et al. Prevention of pneumococcal diseases in the post-seven valent vaccine era: a European perspective. BMC Infect. Dis. 12(1), 207 (2012).[CrossRef] [Medline]
12. Brugger SD, Frey P, Aebi S et al. Multiple colonization with S. pneumoniae before and after introduction of the seven-valent conjugated pneumococcal polysaccharide vaccine. PLoS ONE5(7), e11638 (2010). [CrossRef] [Medline]
13. Strutton DR, Farkouh RA, Earnshaw SR et al. Cost–effectiveness of 13-valent pneumococcal conjugate vaccine: Germany, Greece, and The Netherlands. J. Infect. 64(1), 54–67 (2012).[CrossRef] [Medline]
14. Hanquet G, Perrocheau A, Kissling E et al. Surveillance of invasive pneumococcal disease in 30 EU countries: towards a European system? Vaccine 28(23), 3920–3928 (2010). [CrossRef][Medline]
15. Ansaldi F, De Florentiis D, Canepa P et al. Carriage of Streptoccoccus pneumoniae 7 years after implementation of vaccination program in a population with very high and long-lasting coverage, Italy. Vaccine 30(13), 2288–2294 (2012). [CrossRef] [Medline]
16. Goossens H, Ferech M, Vander Stichele R et al. Outpatient antibiotic use in Europe and association with resistance: a cross-national database study. Lancet 365(9459), 579–587(2005). [CrossRef] [Medline]
17. Canet JJ, Garau J. Importance of dose and duration of beta-lactam therapy in nasopharyngeal colonization with resistant pneumococci. J. Antimicrob. Chemother. 50(Suppl. S2), 39–43(2002). [CrossRef] [Medline] [CAS]
18. Soriano F, Rodriguez-Cerrato V. Pharmacodynamic and kinetic basis for the selection of pneumococcal resistance in the upper respiratory tract. J. Antimicrob. Chemother. 50(Suppl. S2), 51–58 (2002). [CrossRef] [Medline] [CAS]
19. Cornick JE, Bentley SD. Streptococcus pneumoniae: the evolution of antimicrobial resistance to beta-lactams, fluoroquinolones and macrolides. Microbes Infect. 14(7–8), 573–583 (2012).[CrossRef] [Medline] [CAS]
20. Van Bijnen EM, Den Heijer CD, Paget WJ et al. The appropriateness of prescribing antibiotics in the community in Europe: study design. BMC Infect. Dis. 11, 293 (2011). [CrossRef] [Medline]
21. Den Heijer CD, Van Bijnen EM, Paget WJ et al. Prevalence and resistance of commensalStaphylococcus aureus, including meticillin-resistant S aureus, in nine European countries: a cross-sectional study. Lancet Infect. Dis. 13(5), 409–

415 (2013). [CrossRef] [Medline]

22. EUCAST Definitive Document E.DEF 3.1, June 2000: determination of minimum inhibitory concentrations (MICs) of antibacterial agents by agar dilution. Clin. Microbiol. Infect. 6(9),509–515 (2000). [CrossRef] [Medline]
23. Garcia-Rodriguez JA, Fresnadillo Martinez MJ. Dynamics of nasopharyngeal colonization by potential respiratory pathogens. J. Antimicrob. Chemother. 50(Suppl. S2), 59–73 (2002).[CrossRef] [Medline] [CAS]
24. Consortium VI. Impact of childhood pneumococcal vaccination programmes and activities for pneumococcal vaccines in the EU and EEA/EFTA countries (2012).http://venice.cineca.org/VENICE_Survey_PNC_1_2012-02-24.pdf
25. Van Hoek AJ, Sheppard CL, Andrews NJ et al. Pneumococcal carriage in children and adults two years after introduction of the thirteen valent pneumococcal conjugate vaccine in England.Vaccine 32(34), 4349–4355 (2014). [CrossRef] [Medline]
26. Principi N, Terranova L, Zampiero A et al. Pharyngeal colonization by Streptococcus pneumoniae in older children and adolescents in a geographical area characterized by relatively limited pneumococcal vaccination coverage. Pediatr. Infect. Dis. J. 34(4), 426–432 (2015).[CrossRef] [Medline]
27. Diavatopoulos DA, Short KR, Price JT et al. Influenza A virus facilitates Streptococcus pneumoniae transmission and disease. FASEB J. 24(6), 1789–1798 (2010). [CrossRef] [Medline][CAS]
28. Mccullers JA, Mcauley JL, Browall S et al. Influenza enhances susceptibility to natural acquisition of and disease due to Streptococcus pneumoniae in ferrets. J. Infect. Dis. 202(8),1287–1295 (2010). [CrossRef] [Medline]
29. European Centre for Disease Prevention and Control. Antimicrobial consumption interactive database: ESAC-Net (2010).
30. Adriaenssens N, Coenen S, Versporten A et al. European Surveillance of Antimicrobial Consumption (ESAC): outpatient macrolide, lincosamide and streptogramin (MLS) use in Europe (1997–2009). J. Antimicrob. Chemother. 66(Suppl. 6), vi37–vi45 (2011). [Medline] [CAS]
31. Albrich WC, Monnet DL, Harbarth S. Antibiotic selection pressure and resistance inStreptococcus pneumoniae and Streptococcus pyogenes. Emerg. Infect. Dis. 10(3), 514–517(2004). [CrossRef] [Medline]
32. Baquero F, Baquero-Artigao G, Canton R et al. Antibiotic consumption and resistance selection in Streptococcus pneumoniae. J. Antimicrob. Chemother. 50(Suppl. S2), 27–37 (2002).[CrossRef] [Medline] [CAS]
33. Leclercq R, Courvalin P. Resistance to macrolides and related antibiotics in Streptococcus pneumoniae. Antimicrob. Agents Chemother. 46(9), 2727–2734 (2002). [CrossRef] [Medline][CAS]
34. Wise R, Hart T, Cars O et al. Antimicrobial resistance. Is a major threat to public health. BMJ317(7159), 609–610 (1998). [CrossRef] [Medline] [CAS]

TABLES AND FIGURES

Table 1. Baseline characteristics of the participating patients.

Country	Total patient population (n)	Excluded from analysis			Included (n)	Women (% of total)	Age distribution (% of total)				
		Age (n)	Mismatch (n)	Incorrect sampling (n)			4–9	10–17	18–29	30–60	>60
Austria	3380	4	56	11	3309	56.6	1.2	2.2	15.9	50.6	30.0
Belgium	3132	6	101	0	3025	54.4	1.3	3.1	8.2	41.0	46.5
Croatia	4013	1	35	25	3952	59.5	6.0	8.3	9.2	38.9	37.5
France	3870	0	12	1	3857	55	3.4	4.7	11.5	45.4	35.1
Hungary	4017	3	17	165	3832	55.1	8.6	15.7	9.6	36.8	29.4
The Netherlands	3873	14	12	0	3847	57.2	3.2	5.3	11.2	46.1	34.2
Spain	4001	0	11	21	3969	58.4	4.9	5.9	9.2	41.5	38.5
Sweden	3273	33	26	0	3214	57.5	5.4	5.4	9.7	37.9	41.7
UK	3211	3	52	0	3156	59.2	0.0	0.0	14.8	46.4	38.9
Total	32,770	64	322	223	32,161	57	3.9	5.8	10.9	42.6	36.7

Table 2. *Streptococcus pneumoniae* nasal carriage prevalence.

Country	Total swabs (n)	Pneumococcal prevalence			
		Unadjusted (years)		Adjusted (age, gender and GP); (years)	
		4–9 (%)	>10 (%)	4–9 (%)	>10 (%)
Austria	3380	21.5 (13.7–32.2)	1.4 (1.0–2.1)	13.8 (8.6–21.5)	1.3 (0.8–1.7)
Belgium	3132	20.0 (12.8–29.8)	1.5 (1.0–2.3)	12.8 (8.0–19.8)	1.3 (0.9–1.9)
Croatia	4013	23.1 (16.3–31.6)	2.1 (1.4–3.1)	14.9 (10.0–21.4)	1.8 (1.2–2.6)
France	3870	32.0 (23.6–41.8)	3.4 (2.3–5.0)	22.2 (15.7–30.5)	2.8 (1.9–4.0)
Hungary	4017	24.8 (18.0–33.1)	1.5 (1.0–2.3)	17.3 (12.0–24.3)	1.2 (0.8–1.7)
The Netherlands	3873	27.5 (19.6–37.0)	2.5 (1.7–3.6)	16.9 (11.6–23.9)	2.1 (1.4–2.9)
Spain	4001	38.6 (29.5–48.7)	2.9 (2.0–4.2)	24.0 (16.9–32.9)	2.2 (1.5–3.2)
Sweden	3273	27.4 (19.8–36.5)	2.0 (1.3–2.9)	15.3 (10.5–21.7)	1.5 (1.0–2.3)
UK	3211	No data†	1.8 (1.0–2.2)	No data†	1.2 (0.8–1.8)

†All participants were ≥18 years.
GP: General practitioner.

Table 3. Pneumococcal vaccination status in participating patients.

Country	Patients with known vaccination status			Vaccinated patients		
	Total (n)	4–9 years (n)	>10 years (n)	4–9 years % (95% CI)	>10 years % (95% CI)	Total % (95% CI)
Austria	2426	34	2392	8.8 (–0.7–18.3)	7.9 (6.8–9)	7.9 (6.8–9)
Belgium	2942	36	2906	11.1 (0.8–21.4)	16.8 (15.4–18.2)	16.7 (15.4–18)
Croatia	3778	216	3562	3.2 (0.9–5.6)	0.6 (0.4–0.9)	0.7 (0.4–1)
France	3552	122	3430	46.7 (37.9–55.6)	4.1 (3.4–4.8)	5.5 (4.8–6.3)
Hungary	3595	313	3282	7.7 (4.8–10.7)	3.4 (2.8–4)	3.7 (3–4.3)
The Netherlands	3080	97	2983	14.4 (7.4–21.4)	2.5 (1.9–3.1)	2.9 (2.3–3.5)
Spain	3800	192	3608	46.4 (39.4–53.5)	28.3 (26.8–29.8)	29.2 (27.8–30.7)
Sweden	2708	155	2553	5.2 (1.7–8.7)	11.5 (10.3–12.7)	11.2 (10–12.4)
UK	2713	No data†	2713	No data†	28.3 (26.6–30)	28.3 (26.6–30)
Total	28,594	1165	27,449	17.7 (15.5–19.9)	11.3 (10.9–11.7)	11.6 (11.2–12.0)

†All participants were ≥18 years.

Table 4. Lowest and highest prevalence of antibiotic resistance of commensal *S. pneumoniae*.

Antimicrobial agent	Lowest antimicrobial resistance		Highest antimicrobial resistance	
	Prevalence % (95% CI)	Country	Prevalence % (95% CI)	Country
Cefaclor	30.6 (15.6–45.7)	UK	77.3 (64.9–89.7)	Belgium
Cefuroxime	3.9 (0.5–7.2)	The Netherlands	31.1 (24.1–38.1)	Spain
Ceftazidime	2.9 (0.0–6.1)	Sweden	24.6 (17.3–31.9)	Croatia
Clarythromycin	1.9 (0.0–4.5)	Sweden	29.3 (22.4–36.4)	Spain
Clindamycin	1.9 (0.0–4.5)	Sweden	27.5 (20.7–34.3)	Spain
Penicillin	3.9 (0.2–7.6)	Sweden	31.7 (24.6–36.8)	Spain
Tetracyclin	1.9 (0.0–4.5)	Sweden	29.9 (23–36.8)	Spain
Trimethroprim/sulfothoxazole	1.9 (0.0–4.5)	Sweden	28.7 (21.8–35.6)	Spain

No resistance was found to moxiflxacin and ciprofloxacin.

Table 5. Antibiotic resistance prevalence in the nine participating countries.

Country	n	Cefaclor % (95% CI)	Cefuroxime % (95% CI)	Ceftazidime % (95% CI)	Clarythromycin % (95% CI)	Clindamycin % (95% CI)	Penicillin % (95% CI)	Tetracycline % (95% CI)	Trimethroprim-Sulfothoxazole % (95% CI)
Austria	38	36.8 (21.5–52.1)	7.9 (–0.7–16.5)	7.9 (–0.7–16.5)	10.5 (0.8–20.3)	2.6 (–2.5–7.7)	7.9 (–0.7–16.5)	2.6 (–2.5–7.7)	5.3 (–1.8–12.4)
Belgium	44	77.3 (64.9–89.7)	25 (12.2–37.8)	22.7 (10.3–35)	22.7 (10.3–35)	18.2 (6.8–29.6)	25 (12.2–37.8)	20.5 (8.6–32.4)	20.5 (8.6–32.4)
Croatia	134	59.7 (51.4–68.0)	26.9 (19.4–34.4)	24.6 (17.3–31.9)	20.9 (14.0–27.8)	15.7 (9.5–21.9)	22.4 (15.3–29.5)	16.4 (10.1–22.7)	15.7 (9.5–21.9)
France	170	54.1 (46.6–61.6)	25.9 (19.3–32.5)	21.8 (15.6–28)	24.1 (17.7–30.5)	21.2 (15–27.3)	22.9 (16.6–29.2)	20.0 (14–26)	8.8 (4.5–13)
Hungary	116	56.0 (47.0–65.0)	29.3 (21.0–37.6)	22.4 (14.8–30.0)	15.5 (8.9–22.1)	9.5 (4.2–14.8)	26.7 (18.7–34.8)	13.8 (7.5–20.1)	25.9 (17.9–33.9)
The Netherlands	129	34.1 (25.9–42.3)	3.9 (0.5–7.2)	3.9 (0.5–7.2)	3.9 (0.5–7.2)	3.1 (0.1–6.1)	4.7 (1.1–8.4)	3.1 (0.1–6.1)	7.0 (2.6–11.4)
Spain	167	59.9 (52.5–67.4)	31.1 (24.1–38.1)	24.0 (17.5–30.5)	29.3 (22.4–36.4)	27.5 (20.7–34.3)	31.7 (24.6–38.8)	29.9 (23.0–36.8)	28.7 (21.8–35.6)
Sweden	103	48.5 (38.9–58.2)	5.8 (1.3–10.3)	2.9 (0.0–6.1)	1.9 (0.0–4.5)	1.9 (0.0–4.5)	3.9 (0.2–7.6)	1.9 (0.0–4.5)	1.9 (0.0–4.5)
UK	36	30.6 (15.6–45.7)	13.9 (2.6–25.2)	13.9 (2.6–25.2)	13.9 (2.6–25.2)	11.1 (0.9–21.4)	13.9 (2.6–25.2)	11.1 (0.8–21.4)	13.9 (2.6–25.2)
Total	931	52.6 (49.1–55.5)	20.9 (18.3–23.5)	17.3 (14.9–19.7)	17.3 (14.9–19.7)	14.2 (12.0–16.3)	19.4 (16.9–21.9)	15.2 (12.9–17.5)	15.0 (12.7–17.3)

No resistance was found to moxiflxacin and ciprofloxacin.